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REVIEW

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Biomarkers and acute coronary syndromes: an update

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Biomarkers complement clinical assessment and the 12-lead ECG in the diagnosis, risk stratification, triage, and management of patients with suspected acute coronary syndrome (ACS). While there is broad consensus that cardiac troponin (cTn) I or T is the preferred biomarker in clinical practice, important uncertainties remain regarding the value of high-sensitivity cTn assays, their best clinical use including the most appropriate timing of serial measurements, as well as the added value of other biomarkers reflecting and quantifying other pathophysiological signals including copeptin and natriuretic peptides. This review will address these aspects with a focus on the diagnostic application of biomarkers, as they are associated with immediate therapeutic consequences. In addition, this review will briefly highlight that increased diagnostic accuracy offered by high-sensitivity cTn assays has contributed to improve our understanding of the incidence, pathophysiology, and mortality of the two distinct components currently summarized under the term ACS: acute myocardial infarction and unstable angina.

Keywords Cardiac troponin • Cardiomyocyte injury • Myocardial ischaemia • Diagnosis • Acute myocardial infarction • Copeptin • Natriuretic peptides

Introduction

The two components of the acute coronary syndromes (ACS), acute myocardial infarction (AMI) and unstable angina (UA), are major causes of death and disability worldwide.[1–3] The risk of death and the benefit from early revascularization are highest within the first hours; therefore early diagnosis is critical.[1–4]

Approximately 15–20 million patients per year present to the emergency department (ED) with acute chest pain or other symptoms suggestive of ACS in Europe and the USA. As about two-thirds of these patients will be found not to have ACS, rule-in and rule-out seem to be equally important.[1–4]

Current standard of care

Biomarkers complement clinical assessment and the 12-lead ECG in the diagnosis, risk stratification, triage, and management of patients with suspected ACS. While both clinical assessment including a detailed evaluation of chest pain characteristics and the 12-lead ECG are indispensable tools, on their own, they have insufficient accuracy. Therefore, measurement of a biomarker reflecting and quantifying cardiomyocyte injury, preferably cardiac troponin (cTn) I or T, is mandatory in all patients presenting with suspected ACS. The universal definition of myocardial infarction, which is supported by the European Society of Cardiology, the American Heart Association, the American College of Cardiology, and the World Heart Federation, since 2000 has defined the 99th percentile of healthy individuals as the cTn decision value for AMI.[3] Despite this, many institutions worldwide ignore this guideline and continue to use higher cut-off values for cTn.[4] This is of major concern, as this practice incorrectly assigns patients with small AMIs or patients with large AMIs presenting early 'normal' cTn levels and thereby contributes to misdiagnosis and ultimately harms patients.[1–4]

Particularly when discussing the clinical value of biomarkers, it is important to reiterate that ACS includes two distinct clinical entities, AMI (ischaemic chest pain at rest with cardiomyocyte necrosis) and UA (ischaemic chest pain at rest or minimal exertion without cardiomyocyte necrosis), that differ substantially in their risk to the patient and the documented benefit of early revascularization.[1–3]

Cardiac troponin

Cardiac troponin I and T are proteins unique to the heart and specific and sensitive biomarkers of myocardial damage.[2–4] The cTn

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complex is immobilized on the thin filament of the contractile apparatus and plays a critical role in the regulation of excitation–contraction coupling in the heart.[1–3] In AMI, cTnI and cTnT are released from necrotic myocardium both as intact proteins and degradation products. Detection of cTn in peripheral blood indicates and quantifies cardiomyocyte damage. A major limitation of conventional cTn assays is their low sensitivity at the time of patient presentation, owing to a delayed increase in circulating levels requiring serial sampling for 6–9 h in a significant number of patients.[1–6]

Sensitive and high-sensitivity cardiac troponin assays, and the 99th percentile

Recent advances in assay technology have lead to a refinement in cTn assays and therefore the clinical ability to detect and quantify cardiomyocyte injury.[1–4] Sensitive (detection of cTn in ~20–50% of healthy individuals) and high-sensitivity cTn (hs-cTn, detection of cTn in ~50–90% of healthy individuals) assays have two differentiating features from conventional cTn assays: (i) detection of cTn in a substantial number of healthy persons and (ii) a more precise definition of what is 'normal' (= the 99th percentile) with a precision of the assay expressed as the coefficient of variability which should be <20% and preferably <10%.[1–24] These features are of key importance as a cTn value above the 99th percentile of a normal reference population is a 'condition sine qua non' for the diagnosis of AMI. The vast majority of cTn assays run on automated laboratory platforms in Europe are sensitive cTn or hs-cTn, while, e.g. the fourth generation cTnT, a conventional cTn assay, is still used in many institutions in the USA.[4,19,24] In addition, many point-of care assays used worldwide also cannot be considered sensitive or high-sensitivity.[21–24]

Data from several large multicentre studies have consistently shown that sensitive cTn and hs-cTn assays increase the accuracy of the diagnosis of AMI at the time of presentation to the ED.[7–23] The benefit observed for sensitive cTn and hs-cTn assays was most pronounced in patients presenting early after chest pain onset and could be established also in challenging subgroups such as elderly patients or patients with pre-existing coronary artery disease.[12,13] The use of sensitive cTn or hs-cTn assays allows a more rapid rule-in and rule-out. While both sensitive cTn and hs-cTn assays are clearly superior to conventional cTn assays, it is currently still unknown and a topic of on-going research whether and to what extent the use of hs-cTn assays provides a clinically meaningful advantage with compared with the use of sensitive cTn assays.

Concerns and pitfalls with high-sensitivity cardiac troponin

While hs-cTn assays have been introduced into clinical practice in Europe and many other countries (hs-cTnT, Roche in 2010; hs-cTnI, Abbott in 2013), these assays still await approval for clinical use in the USA. Most concerns regarding the introduction of hs-cTn assays including the increased detection of patients with cardiomyocyte damage from causes other than AMI are in fact concerns to use the 99th percentile.[2–5,23,24]

It is important to highlight a potential iatrogenic pitfall: levels of cTn have been reported in μg/L in the last two decades. Despite certain analytical differences among cTn assays, cardiologists and ED physicians have learned that cTn levels of e.g. 5 or 10 μg/L occur very rarely and are almost always due to very large AMIs. Now experts have begun to advocate to report levels of cTn if measured with a hs-cTn assay in ng/L in order to avoid errors associated with decimals.[2,19] This would mean that a normal level of hs-cTn would be reported as e.g. 5 ng/L rather than 0.005 μg/L and a large AMI would be reported as e.g. 5000 ng/L rather than 5 μg/L. This transition obviously requires extensive communication between laboratory experts and clinicians in each hospital in order to avoid iatrogenic harm due to incorrect interpretation of hs-cTn results.

High-sensitivity cardiac troponin: timing of serial measurements and the ESC guidelines

Due to the higher sensitivity for the detection of AMI at presentation, the time interval to the second measurement of hs-cTn can be significantly shortened with the use of hs-cTn assays (*Figures 1* and *2*).[1–5] This is the main clinical advantage of these assays and may result in substantial reduction in time to decision and therefore total treatment costs in the ED.[1–5] While the ESC guidelines use the term hs-cTn when referring to the option of using a 3 h rule-out protocol,[5] it is very likely that the 3 h rule-out protocol also can be safely applied with at least some sensitive cTn assays, as the reference study used to document the safety of the novel approach used a sensitive cTn (not a hs-cTn) assay.[15] As shown in *Figure 1*, the 3 h rule-out protocol is not based on hs-cTn levels only, but also requires patients to be pain-free and to have a GRACE score below 140.[5] In addition, the ESC guidelines for the first time highlight the time since chest pain onset as a modifier. The onset of chest pain in patients with AMI is considered the onset of cardiomyocyte damage and therefore the release of cTn into the circulation. In patients who provide a precise estimate of the onset of acute chest pain, the time interval from chest pain onset to blood sampling can help interpret cTn findings: AMI may be safely ruled out without a second measurement once hs-cTn levels are normal in a patient presenting with a chest pain onset >6 h prior to ED presentation, so the second cTn draw could be restricted to patients with chest pain onset <6 h or patients in whom this information cannot be reliably obtained.[5] Although this concept is very appealing, it should only be applied cautiously and still requires prospective testing. Regarding the rule-in of AMI, a significant rise and/or fall in cTn is an important criterium to differentiate AMI from chronic causes of cardiomyocyte injury.[2,5]

Rapid rule-out and rule-in protocols

It is a matter of on-going research whether it is possible to safely and reliable rule-out and rule-in AMI even earlier as the ESC 3 h rule-out protocol.[25–35] Data from large observational multicentre studies showed an excellent performance of 2 h rule-out protocols that combine hs-cTn values with clinical information,[7–9] but also for a 1 h rule-out and rule-in protocol exclusively based on hs-cTnT values.[10] This approach maximizes the use of the diagnostic

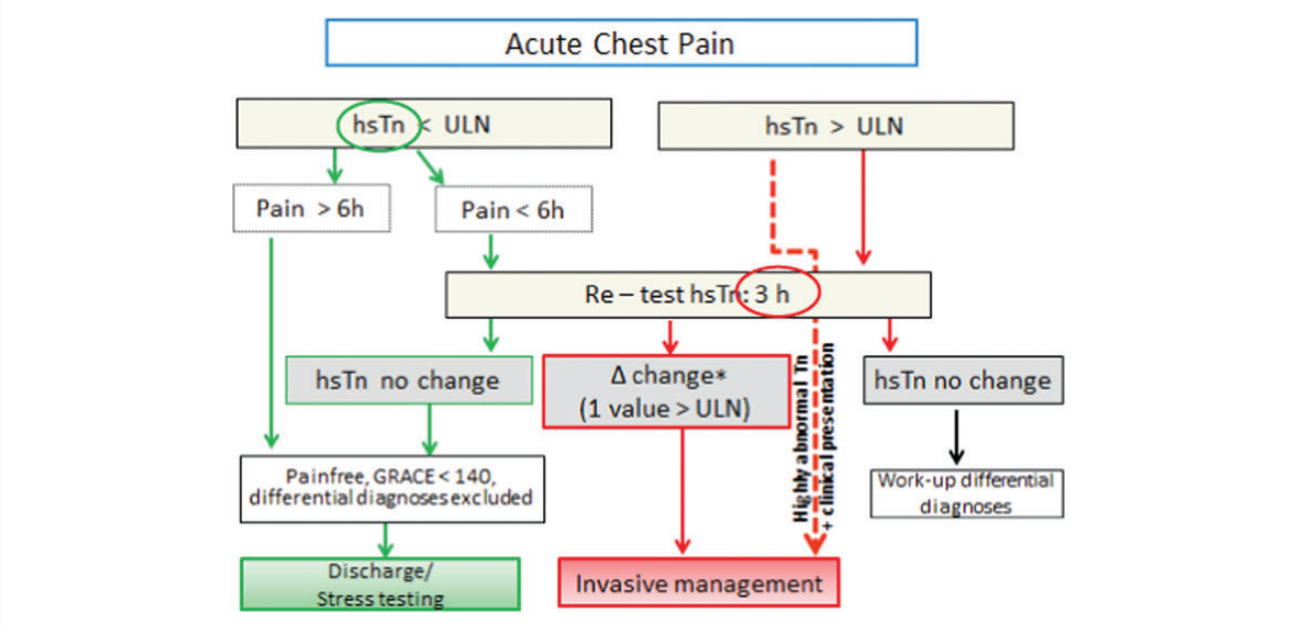

Figure 1 Algorithm for the use of high-sensitivity cardiac troponin levels suggested in the 2011 ESC NSTEMI guidelines (adopted from ref. 3).

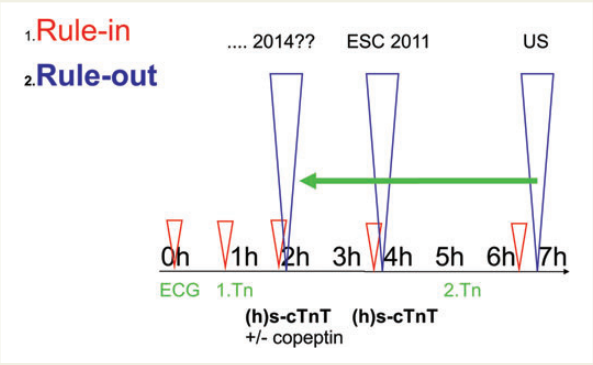

Figure 2 The use of sensitive or high-sensitivity cardiac troponin ((h)s-cTn) allows more rapid rule-out and more rapid rule-in. Rule-in of acute myocardial infarction (AMI) can be at presentation (0 h) in patients with unequivocal ST-elevations, at 1 h in patients with elevations in cardiac troponin (cTn) in the measurement performed at presentation (turn-around time is around 1 h in most hospitals) but only at 7 h if the first cTn is normal and the elevation in cTn becomes apparent only at the second measurement performed after 6 h. Rule-out requires a normal second cTn level and therefore 7 h. According to the 2011 ESC NSTEMI guidelines, AMI can be reliably ruled-out at 4 h if a (h)s-cTn assay is used. Recent research has indicated that (h)s-cTn assays may allow even earlier rule-out and rule-in if assay-specific algorithms are used or if applied in combination with copeptin.

information included in hs-cTn values by combining two different assay-specific cut-off values for rule-out and rule-in with absolute changes within the first hour after presentation.[22] This innovative approach should only be considered for clinical use after successful external validation. Further highlighting the diagnostic value of interpreting hs-cTn concentrations as quantitative values (*Figure 3*),[28] data from two independent large studies have shown that very low hs-cTn levels (e.g. levels below the limit of detection of the hs-cTnT assays on their own) have a very high negative predictive value of about 99% for the presence of AMI and may obviate the need for serial testing in selected patients.[24,26,33]

Unstable angina in the high-sensitivity cardiac troponin era

Some expert had hypothesized that the diagnosis of UA would disappear with the clinical introduction of hs-cTn assays.[2] While the condition currently termed UA may not disappear, the available data suggest that our current classification scheme that unifies UA together with AMI as ACS should be reconsidered. By increasing the detection of patients with small AMIs which were previously incorrectly classified as UA, the clinical introduction of hs-cTn assays results in a modest increase in the incidence of AMI and a reciprocal modest decrease in the incidence of UA.[27] In marked contrast to patients with AMI and similar to patients with non-cardiac causes of acute chest pain, most patients with UA do not exhibit hs-cTn changes (*Figure 4*).[29] This indicates the absence of distal coronary embolization of the culprit lesion as a key differentiating pathophysiological feature between UA and AMI. When compared with NSTEMI, also the risk of death is substantially lower in UA, as seem to be the benefit from early revascularization and aggressive antiplatelet therapy.[30] These observations combined suggest that in the hs-cTn era UA may well be better placed as a subgroup of severe stable CAD rather than together with AMI.[27–30]

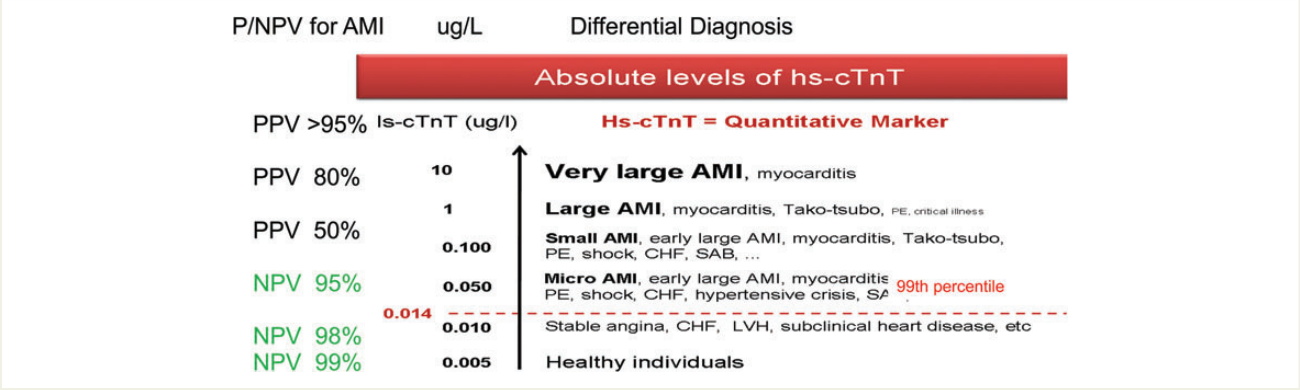

Figure 3 Cardiac troponin (cTn) should be interpreted as a quantitative marker of cardiomyocyte injury. In patients presenting with acute chest pain, the lower the level, the lower the likelihood and the higher the negative predictive value (NPV) for the presence of acute myocardial infarction (AMI). Thereby, the differential diagnosis is broad. In contrast, the higher the level, the higher the likelihood and the higher the positive predictive value (PPV) for the presence of AMI. The differential diagnosis is much smaller. Importantly, mild elevations of cTn with levels just above the 99th percentile have a broad differential diagnosis and therefore a rather low PPV for AMI of only ~50%. Levels of hs-cTnT are given as μg/L.

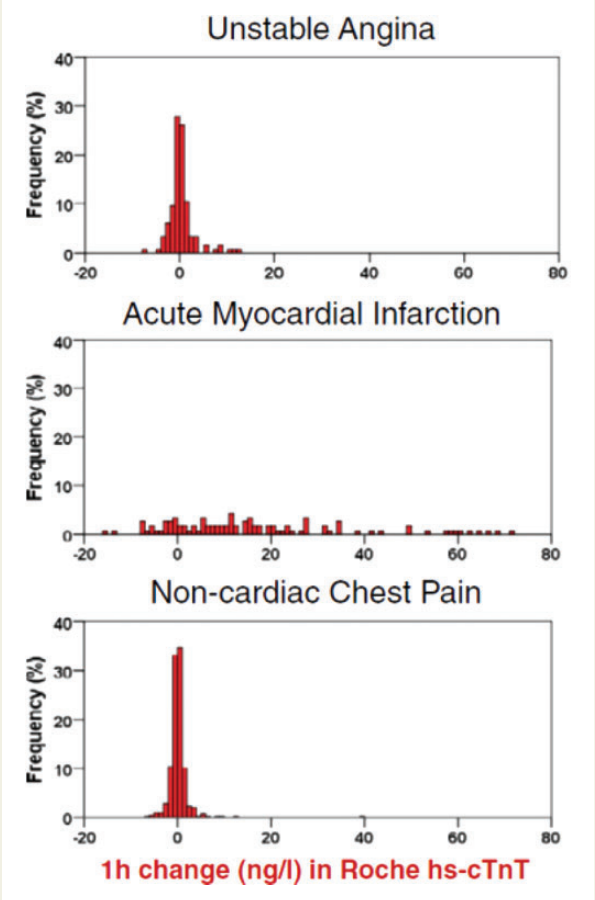

Figure 4 Changes of high-sensitivity cardiac troponin within the first hour after presentation according to the adjudicated final diagnosis as surrogate measures to quantify the extent of acute cardiomyocyte injury that occurred in the preceding hours. Frequency histograms of changes within the first hour in ng/L for Roche high-sensitivity cardiac troponin T. Adapted from ref. 29.

Alternative biomarkers reflecting cardiomyocyte damage: heart-type fatty-acid binding protein

Heart-type fatty acid-binding protein (h-FABP), a small soluble cytosolic protein involved in the transportation of long-chain fatty acids into the cardiomyocyte, is released rapidly into the circulation in response to cardiomyocyte injury. Due to its solubility and small size (15 kDa), h-FABP can be released more rapidly than structurally bound molecules like cTns.

Thus, h-FABP is regarded as an early sensitive marker of AMI.[35–37] Most of the promising data regarding the potential clinical value of h-FABP were obtained prior the clinical introduction of hs-cTn assays. When used on top of hs-cTnT, h-FABP did not seem not provide relevant added diagnostic value in consecutive patients. Ongoing research is therefore focusing on the use of h-FABP in patients presenting very early (<1–2 h since chest pain onset).

Biomarkers reflecting endogenous stress: copeptin

Copeptin, the c-terminal part of the vasopressin prohormone, is secreted stochiometrically with arginin–vasopressin from the neurohypophysis and seems to quantify the individual endogenous stress level and also the mortality risk in multiple medical conditions including AMI.[31–34] As endogenous stress is present already at the onset of AMI, copeptin appears to be able to identify AMI very early after symptom onset, even when cTn is still negative (*Figure 5*).[31–34,38–41] As the time course of endogenous stress and detectable cardiomyocyte damage seems to be reciprocal, copeptin seems to be the ideal marker to compensate for the sensitivity deficit of conventional cTn assays in early presenters. When used in conjunction with conventional fourth generation cTnT, the added value of copeptin regarding diagnostic accuracy already at presentation was substantial.[31,32] The added value of copeptin when used

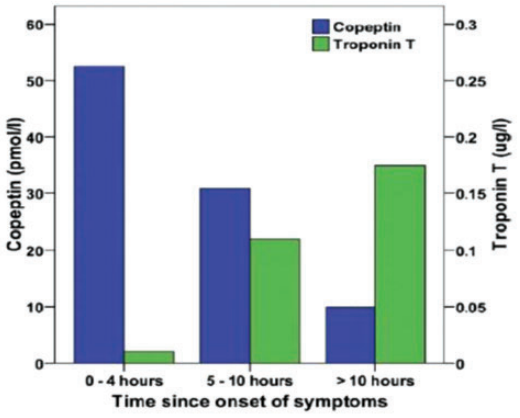

Figure 2 Copeptin and Troponin T Levels at Presentation in Relation to Time Since Onset of Symptoms

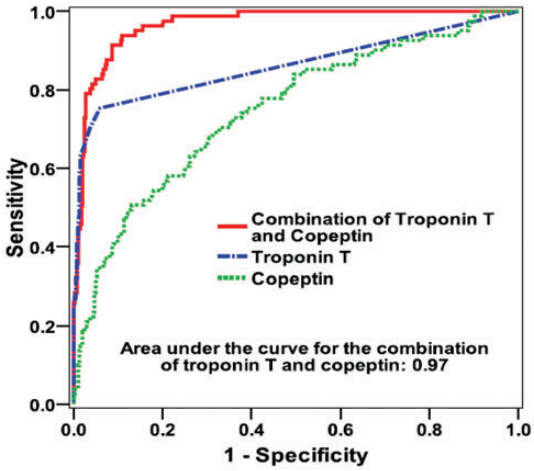

Figure 4 ROC Curves at Presentation for the Diagnosis of AMI

Figure 5 Incremental value of copeptin when used in combination with the fourth generation cardiac troponin T (cTnT) assay: combining two different pathophysiological signals in the early diagnosis of acute myocardial infarction. Left: levels of copeptin and cTnT according to the time since the onset of acute chest pain in patients with acute myocardial infarction. As the time course of endogenous stress and detectable cardiomyocyte damage seems to be reciprocal, copeptin seems to be the ideal marker to compensate for the sensitivity deficit of conventional cTn assays in early presenters. Right: when used in conjunction with conventional fourth generation cTnT, the added value of copeptin regarding diagnostic accuracy at presentation is substantial. Adapted from ref. 31.

in combination with sensitive cTnI or hs-cTnT seems to be smaller.[31–34,38–41] The concept in which copeptin seems to have the greatest appeal to clinicians is its use within a dual-marker strategy for very early rule-out of AMI: Patients with acute chest pain presenting to the ED with initial negative values (below the 99th percentile) of either sensitive cTn or hs-cTn and also low levels of copeptin (e.g. $<$10 pmol/L) do have a very high negative predictive value (around 99%) for AMI and seem to be appropriate candidates for rapid discharge from the ED without the need for serial cTn testing. In fact, a recent large multicentre randomized controlled study evaluating the safety and efficacy of this approach when compared with standard of care (second cTn measurement after 3–6 h) confirmed this concept.[42]

Other complimentary prognostic biomarkers

Natriuretic peptides, midregional pro-adrenomedullin, and GDF-15 are powerful predictors of mortality in patients with suspected or established ACS, but do not seem to provide added diagnostic information.[43–47] It is currently unclear how the pathophysiological signals quantified by these different markers (e.g. haemodynamic cardiac stress in the case of natriuretic peptides) could be best used clinically, in order to mitigate the high mortality risk identified in patients with high levels of these markers.

Biomarkers associated with plaque instability

As inflammation plays a key role in atherosclerotic plaque formation, plaque destabilization, and plaque disruption, and in ACS plaque rupture invariably precedes cardiomyocyte damage by minutes to potentially hours, biomarkers of plaque instability are logical candidates for early AMI markers.[48,49] Unfortunately, recent studies have consistently shown that at least when measured with currently available assays markers of plaque instability including myeloperoxidase, myeloid-related protein 8/14, pregnancy-associated plasma protein-A, and C-reactive protein have very low diagnostic accuracy and therefore are not helpful in the early diagnosis of AMI.[48,49]

In conclusion, hs-cTn measured at presentation and after 3 h form the new standard of care since 2011. Likely, even more rapid hs-cTn protocols with or without the additional use of copeptin will be the strategies of the future.

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References

1. Thygesen K, Mair J, Katus H, Plebani M, Venge P, Collinson P, Lindahl B, Giannitsis E, Hasin Y, Galvani M, Tubaro M, Alpert JS, Biasucci LM, Koenig W, Mueller C, Huber K, Hamm C, Jaffe AS. Recommendations for the use of cardiac troponin measurement in acute cardiac care. *Eur Heart J* 2010;**31**:2197–2204.
2. Thygesen K, Mair J, Giannitsis E, Mueller C, Lindahl B, Blankenberg S, Huber K, Plebani M, Biasucci LM, Tubaro M, Collinson P, Venge P, Hasin Y, Galvani M, Koenig W, Hamm C, Alpert JS, Katus H, Jaffe AS. the Study Group on Biomarkers in Cardiology of the ESCWGoACC. How to use high-sensitivity cardiac troponins in acute cardiac care. *Eur Heart J* 2012;**33**:2252–2257.
3. Thygesen K, Alpert JS, Jaffe AS, Simoons ML, Chaitman BR, White HD. Joint ESC/ACCF/AHA/WHF Task Force for the Universal Definition of Myocardial. Third universal definition of myocardial infarction. *Eur Heart J* 2012;**33**:2551–2567.

4. Safford MM, Parmar G, Barasch CS, Halanych JH, Glasser SP, Goff DC, Prineas RJ, Brown TM. Hospital laboratory reporting may be a barrier to detection of 'micro-size' myocardial infarction in the US: an observational study. *BMC Health Serv Res* 2013;**13**:162.
5. Hamm CW, Bassand JP, Agewall S, Bax J, Boersma E, Bueno H, Caso P, Dudek D, Gielen S, Huber K, Ohman M, Petrie MC, Sonntag F, Uva MS, Storey RF, Wijns W, Zahger D. ESC Committee for Practice Guidelines. ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation: The Task Force for the management of acute coronary syndromes (ACS) in patients presenting without persistent ST-segment elevation of the European Society of Cardiology (ESC). *Eur Heart J* 2011;**32**:2999–3054.
6. Anderson JL, Adams CD, Antman EM, Bridges CR, Califf RM, Casey DE Jr, Chavey WE 2nd, Fesmire FM, Hochman JS, Levin TN, Lincoff AM, Peterson ED, Theroux P, Wenger NK, Wright RS, Jneid H, Ettinger SM, Ganiats TG, Lincoff AM, Philippides GJ, Zidar JP. American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. 2012 ACCF/AHA focused update incorporated into the ACCF/AHA 2007 guidelines for the management of patients with unstable angina/non-ST-elevation myocardial infarction: a report of the American College of Cardiology Foundation/American Heart Association Task Force on Practice Guidelines. *Circulation* 2013;**127**:e663–e828.
7. Than M, Cullen L, Reid CM, Lim SH, Aldous S, Ardagh MW, Peacock WF, Parsonage WA, Ho HF, Ko HF, Kasliwal RR, Bansal M, Soerianata S, Hu D, Ding R, Hua Q, Seok-Min K, Sritara P, Sae-Lee R, Chiu TF, Tsai KC, Chu FY, Chen WK, Chang WH, Flaws DF, George PM, Richards AM. A 2-h diagnostic protocol to assess patients with chest pain symptoms in the Asia-Pacific region (ASPECT): a prospective observational validation study. *Lancet* 2011;**377**:1077–1084.
8. Than M, Cullen L, Aldous S, Parsonage WA, Reid CM, Greenslade J, Flaws D, Hammett CJ, Beam DM, Ardagh MW, Troughton R, Brown AF, George P, Florkowski CM, Kline JA, Peacock WF, Maisel AS, Lim SH, Lamanna A, Richards AM. 2-Hour accelerated diagnostic protocol to assess patients with chest pain symptoms using contemporary Troponins as the only biomarker. *J Am Coll Cardiol* 2012;**59**:2091–2098.
9. Cullen L, Mueller C, Parsonage WA, Wildi K, Greenslade JH, Twerenbold R, Aldous S, Meller B, Tate JR, Reichlin T, Hammett CJ, Zellweger C, Ungerer Mmed JP, Gimenez MR, Troughton R, Murray K, Brown AF, Mueller M, George P, Mosimann T, Flaws DF, Reiter M, Lamanna A, Haaf P, Pemberton CJ, Richards AM, Chu K, Reid CM, Peacock WF, Jaffe AS, Florkowski C, Deely JM, Than M. Validation of high-sensitivity troponin I in a 2-h diagnostic strategy to assess 30-day outcomes in emergency-department patients with possible acute coronary syndrome. *J Am Coll Cardiol* 2013;**62**:1242–1249.
10. Reichlin T, Schindler C, Drexler B, Twerenbold R, Reiter M, Zellweger C, Moehring B, Ziller R, Hoeller R, Rubini Gimenez M, Haaf P, Potocki M, Wildi K, Balmelli C, Freese M, Stelzig C, Freidank H, Osswald S, Mueller C. One-hour rule-out and rule-in of acute myocardial infarction using high-sensitivity cardiac troponin t. *Arch Intern Med* 2012;**172**:1211–1218.
11. Haaf P, Drexler B, Reichlin T, Twerenbold R, Reiter M, Meissner J, Schaub N, Stelzig C, Freese M, Heinzelmann A, Meune C, Balmelli C, Freidank H, Winkler K, Denhaerynck K, Hochholzer W, Osswald S, Mueller C. High-sensitivity cardiac troponin in the distinction of acute myocardial infarction from acute cardiac noncoronary artery disease. *Circulation* 2012;**126**:31–40.
12. Reiter M, Twerenbold R, Reichlin T, Haaf P, Meissner J, Hochholzer W, Stelzig C, Freese M, Heinisch C, Breidthardt T, Freidank H, Winkler K, Campodarve I, Gea J, Mueller C. Early diagnosis of acute myocardial infarction in the elderly using more sensitive cardiac troponin assays. *Eur Heart J* 2011;**32**:1379–1389.
13. Reiter M, Twerenbold R, Reichlin T, Benz B, Haaf P, Meissner J, Hochholzer W, Stelzig C, Freese M, Heinisch C, Balmelli C, Drexler B, Freidank H, Winkler K, Campodarve I, Gea J, Mueller C. Early diagnosis of acute myocardial infarction in patients with pre-existing coronary artery disease using more sensitive cardiac troponin assays. *Eur Heart J* 2012;**33**:988–997.
14. Reichlin T, Hochholzer W, Bassetti S, Steuer S, Stelzig C, Hartwiger S, Biedert S, Schaub N, Buerge C, Potocki M, Noveanu M, Breidthardt T, Twerenbold R, Winkler K, Bingisser R, Mueller C. Early diagnosis of myocardial infarction with sensitive cardiac troponin assays. *N Engl J Med* 2009;**361**:858–867.
15. Keller T, Zeller T, Peetz D, Tzikas S, Roth A, Czyz E, Bickel C, Baldus S, Warnholtz A, Frohlich M, Sinning CR, Eleftheriadis MS, Wild PS, Schnabel RB, Lubos E, Jachmann N, Genth-Zotz S, Post F, Nicaud V, Tiret L, Lackner KJ, Munzel TF, Blankenberg S. Sensitive troponin I assay in early diagnosis of acute myocardial infarction. *N Engl J Med* 2009;**361**:868–877.
16. Keller T, Zeller T, Ojeda F, Tzikas S, Lillpopp L, Sinning C, Wild P, Genth-Zotz S, Warnholtz A, Giannitsis E, Möckel M, Bickel C, Peetz D, Lackner K, Baldus S, Münzel T, Blankenberg S. Serial changes in highly sensitive troponin I assay and early diagnosis of myocardial infarction. *JAMA* 2011;**306**:2684–2693.
17. Giannitsis E, Kurz K, Hallermayer K, Jarausch J, Jaffe AS, Katus HA. Analytical validation of a high-sensitivity cardiac troponin T assay. *Clin Chem* 2010;**56**:254–261.
18. Keller T, Münzel T, Blankenberg S. Making it more sensitive: the new era of troponin use. *Circulation* 2011;**123**:1361–1363.
19. Korley FK, Jaffe AS. Preparing the United States for high-sensitivity cardiac troponin assays. *J Am Coll Cardiol* 2013;**61**:1753–1758.
20. Mueller C. New ESC guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. *Swiss Med Wkly* 2012;**142**:w13514.
21. Apple FS. A new season for cardiac troponin assays: it's time to keep a scorecard. *Clin Chem* 2009;**55**:1303–1306.
22. Reichlin T, Irfan A, Twerenbold R, Reiter M, Hochholzer W, Burkhalter H, Bassetti S, Steuer S, Winkler K, Peter F, Meissner J, Haaf P, Potocki M, Drexler B, Osswald S, Mueller C. Utility of absolute and relative changes in cardiac troponin concentrations in the early diagnosis of acute myocardial infarction. *Circulation* 2011;**124**:136–145.
23. Newby LK, Jessie RL, Babb JD, Christensen RH, De Fer TM, Diamond GA, Fesmire FM, Gersh BJ, Geraci SA, Larsen GC, Kaul S, McKay CR, Philippides GJ, Weintraub WS. ACCF 2012 expert consensus document on practical clinical considerations in the interpretation of troponin elevations: a report of the American College of Cardiology Foundation Task Force on Clinical Expert Consensus Documents. *J Am Coll Cardiol* 2012;**60**:2419–2455.
24. Rubini Gimenez M, Rubini Giménez M, Hoeller R, Reichlin T, Zellweger C, Twerenbold R, Reiter M, Moehring B, Wildi K, Mosimann T, Mueller M, Meller B, Hochgruber T, Ziller R, Sou SM, Murray K, Sakarikos K, Ernst S, Gea J, Campodarve I, Vilaplana C, Haaf P, Steuer S, Minners J, Osswald S, Mueller C. Undetectable Rapid rule out of acute myocardial infarction using undetectable levels of high-sensitivity cardiac troponin. *Int J Cardiol* 2013 [Epub ahead of print].
25. Hammarsten O, Fu ML, Sigurjonsdottir R, Petzold M, Said L, Landin-Wilhelmsen K, Widgren B, Larsson M, Johanson P. Troponin T percentiles from a random population sample, emergency room patients and patients with myocardial infarction. *Clin Chem* 2012;**58**:628–637.
26. Body R, Carley S, McDowell G, Jaffe AS, France M, Cruickshank K, Wibberley C, Nuttall M, Mackway-Jones K. Rapid exclusion of acute myocardial infarction in patients with undetectable troponin using a high-sensitivity assay. *J Am Coll Cardiol* 2011;**58**:1332–1339.
27. Reichlin T, Twerenbold R, Reiter M, Steuer S, Bassetti S, Balmelli C, Winkler K, Kurz S, Stelzig C, Freese M, Drexler B, Haaf P, Zellweger C, Osswald S, Mueller C. Introduction of high-sensitivity troponin assays: impact on myocardial infarction incidence and prognosis. *Am J Med* 2012;**125**:1205–1213.
28. Twerenbold R, Jaffe AS, Reichlin T, Reiter M, Mueller C. High-sensitive troponin T in acute coronary syndrome: what do we gain and what are the challenges? *Eur Heart J* 2012;**33**:579–586.
29. Reichlin T, Twerenbold R, Maushart C, Reiter M, Moehring B, Schaub N, Balmelli C, Rubini Gimenez M, Hoeller R, Sakarikos K, Drexler B, Haaf P, Osswald S, Mueller C. Risk stratification in patients with unstable angina using absolute serial changes of 3 high-sensitive troponin assays. *Am Heart J* 2013;**165**:371–378.
30. Braunwald E, Morrow DA. Unstable angina. Is it time for a requiem? *Circulation* 2013;**127**:2452–2457.
31. Reichlin T, Hochholzer W, Stelzig C, Laule K, Freidank H, Morgenthaler NG, Bergmann A, Potocki M, Noveanu M, Breidthardt T, Christ A, Boldanova T, Merki R, Schaub N, Bingisser R, Christ M, Mueller C. Incremental value of copeptin for rapid rule out of acute myocardial infarction. *J Am Coll Cardiol* 2009;**54**:60–68.
32. Keller T, Tzikas S, Zeller T, Czyz E, Lillpopp L, Ojeda FM, Roth A, Bickel C, Baldus S, Sinning CR, Wild PS, Lubos E, Peetz D, Kunde J, Hartmann O, Bergmann A, Post F, Lackner KJ, Genth-Zotz S, Nicaud V, Tiret L, Münzel TF, Blankenberg S. Copeptin improves early diagnosis of acute myocardial infarction. *J Am Coll Cardiol* 2010;**55**:2096–2106.
33. Meune C, Balmelli C, Vogler E, Twerenbold R, Reiter M, Reichlin T, Haaf P, Drexler B, Wildi K, Hoeller R, Rubini Gimenez M, Moehring B, Zellweger C, Potocki M, Mueller C. Consideration of high-sensitivity troponin values below the 99th percentile at presentation: does it improve diagnostic accuracy? *Int J Cardiol* 2013 [Epub ahead of print].
34. Giannitsis E, Kehayova T, Vafaie M, Katus HA. Combined testing of high-sensitivity troponin t and copeptin on presentation at prespecified cutoffs improves rapid rule-out of non-ST-segment elevation myocardial infarction. *Clin Chem* 2011;**57**:1452–1455.
35. Bruins Slot MH, Reitsma JB, Rutten FH, Hoes AW, van der Heijden GJ. Heart-type fatty acid-binding protein in the early diagnosis of acute myocardial infarction: a systematic review and meta-analysis. *Heart* 2010;**96**:1957–1963.
36. Kurz K, Giannitsis E, Becker M *et al.* Comparison of the new high sensitive cardiac troponin T with myoglobin, h-FABP and cTnT for early identification of myocardial necrosis in the acute coronary syndrome. *Clin Res Cardiol* 2011;**100**:209–215.
37. Reiter M, Twerenbold R, Reichlin T, Mueller M, Hoeller R, Moehring B, Haaf P, Wildi K, Merk S, Bernhard D, Zellweger Mueller C, Freese M, Freidank H, Campodarve Botet I, Mueller C. Heart-type fatty acid-binding protein in the early diagnosis of acute myocardial infarction. *Heart* 2013;**99**:708–714.

38. Potocki M, Reichlin T, Thalmann S, Zellweger C, Twerenbold R, Reiter M, Steuer S, Bassetti S, Drexler B, Stelzig C, Freese M, Winkler K, Haaf P, Balmelli C, Hochholzer W, Osswald S, Mueller C. Diagnostic and prognostic impact of copeptin and high-sensitivity cardiac troponin T in patients with pre-existing coronary artery disease and suspected acute myocardial infarction. *Heart* 2012;**98**:558–565.
39. Ray P, Charpentier S, Chenevier-Gobeaux C, Reichlin T, Twerenbold R, Claessens YE, Jourdain P, Riou B, Mueller C. Combined copeptin and troponin to rule out myocardial infarction in patients with chest pain and a history of coronary artery disease. *Am J Em Med* 2012;**30**:440–448.
40. Balmelli C, Meune C, Twerenbold R, Reichlin T, Rieder S, Drexler B, Rubini Gimenez M, Mosimann T, Reiter M, Haaf P, Mueller M, Ernst S, Ballarino P, Alafify AA, Zellweger C, Wildi K, Moehring B, Vilaplana C, Bernhard D, Merk S, Ebmeyer S, Freidank H, Osswald S, Mueller C. Comparison of the performances of cardiac troponins, including sensitive assays, and copeptin in the diagnosis of acute myocardial infarction and long-term prognosis between women and men. *Am Heart J* 2013;**166**:30–37.
41. Maisel A, Mueller C, Neath SX, Christenson RH, Morgenthaler NG, Nowak RM, Vilke G, Daniels LB, Hollander JE, Apple FS, Cannon C, Nagurney JT, Schreiber D, Defilippi C, Hogan C, Diercks DB, Stein JC, Headden G, Limkakeng AT Jr, Anand I, Wu AH, Papassotiriou J, Hartmann O, Ebmeyer S, Clopton P, Jaffe AS, Frank Peacock W. Copeptin helps 'Copeptin Helps in the Early Detection Of Patients with Acute Myocardial Infarction': the primary results of the CHOPIN Trial. *J Am Coll Cardiol* 2013;**62**:150–160.
42. Moeckel M. Instant early rule-out using cardiac troponin and copeptin in low- to intermediate-risk patients with suspected ACS: a prospective, randomized multicenter study for early discharge. Hotline Session of the ESC Meeting, 3 September, Amsterdam, 2013.
43. Haaf P, Twerenbold R, Reichlin T, Faoro J, Reiter M, Meune C, Steuer S, Bassetti S, Ziller R, Balmelli C, Campodarve I, Zellweger C, Kilchenmann A, Irfan A, Papassotiriou J, Drexler B, Mueller C. Mid-regional pro-adrenomedullin in the early evaluation of acute chest pain patients. *Int J Cardiol* 2012.
44. Khan SQ, O'Brien RJ, Struck J, Quinn P, Morgenthaler N, Squire I, Davies J, Bergmann A, Ng LL. Prognostic value of midregional proadrenomedullin in patients with acute myocardial infarction: the LAMP (Leicester Acute Myocardial Infarction Peptide) study. *J Am Coll Cardiol* 2007;**49**:1525–1532.
45. Haaf P, Reichlin T, Corson N, Twerenbold R, Reiter M, Steuer S, Bassetti S, Winkler K, Stelzig S, Heinisch H, Drexler B, Freidank H, Mueller C. B-type natriuretic peptide in the early diagnosis and risk stratification of acute chest pain. *Am J Med* 2011;**124**:444–452.
46. Widera C, Pencina MJ, Meisner A, Kempf T, Bethmann K, Marquardt I, Katus HA, Giannitsis E, Wollert KC. Adjustment of the GRACE score by growth differentiation factor 15 enables a more accurate appreciation of risk in non-ST-elevation acute coronary syndrome. *Eur Heart J* 2012;**33**:1095–1104.
47. Meune C, Twerenbold R, Drexler B, Balmelli C, Wolf C, Haaf P, Reichlin R, Irfan A, Reiter M, Zellweger C, Meissner J, Stelzig C, Freese M, Capodarve I, Mueller C. Mid-regional pro-A-type natriuretic peptide for diagnosis and prognosis in patients with suspected acute myocardial infarction. *Am J Cardiol* 2012;**109**:1117–1123.
48. Altwegg LA, Neidhart M, Hersberger M, Muller S, Eberli FR, Corti R, Roffi M, Sütsch G, Gay S, von Eckardstein A, Wischnewsky MB, Lüscher TF, Maier W. Myeloid-related protein 8/14 complex is released by monocytes and granulocytes at the site of coronary occlusion: a novel, early, and sensitive marker of acute coronary syndromes. *Eur Heart J* 2007;**28**:941–948.
49. Schaub N, Reichlin T, Meune C, Twerenbold R, Haaf P, Hochholzer W, Niederhauser N, Bosshard P, Stelzig C, Freese M, Reiter M, Gea J, Buser A, Mebazaa A, Osswald S, Mueller C. Markers of plaque instability in the early diagnosis and risk stratification of acute myocardial infarction. *Clin Chem* 2012;**58**:246–256.